FINAL REPORT FY2015
HABITAT ASSESSMENT FUNDED RESEARCH

Project Title: Estimating habitat-specific variability in growth rates of juvenile penaeid shrimps for incorporation into stock assessment models

Funding Fiscal Year: FY2014
Report Fiscal Year and Quarter: FY2015-Q4
Date Submitted: 09/28/2015

Principal Investigator(s): Lawrence Rozas[1], Thomas Minello[2], Jennifer Doerr[2], and Rick Hart[2], [1]NOAA Fisheries, SEFSC/EHCFC, Lafayette, LA; [2]NOAA Fisheries, SEFSC Galveston Laboratory, Galveston, TX

Goals:

The main goal of our project was to estimate and compare growth rates of juvenile white shrimp *Litopenaeus setiferus* and juvenile brown shrimp *Farfantepenaeus aztecus* between vegetated (*Spartina alterniflora*) salt marsh and shallow nonvegetated bottom (SNB). Field experiments and laboratory microcosm experiments were used to estimate shrimp growth rates in both habitat types (marsh and SNB). The field mesocosm experiments were conducted in two estuaries of the northern Gulf of Mexico (EGB = East Galveston Bay and SL = Sabine Lake) to examine between-estuary variation in shrimp growth rates. A second goal of this project was to incorporate this information on growth rates into the new SS-3 assessment model for these species.

Approach:

Field Experiments

We used two different methods to estimate growth rates in the field. In the first approach, we used 10 mesocosms placed in each habitat type (marsh and SNB) within and around the edge of a marsh pond at each location. Mesocosms were 1.07-m in diameter, constructed of 3.2-mm mesh nylon netting, and enclosed 0.89 m^2 of habitat (Rozas & Minello 2011). These experiments, used to estimate growth rates of juvenile white shrimp, were conducted in EGB August 18-25, 2014 and in SL August 29 – September 4, 2014. At each location, the intertidal creek leading into the experimental pond was partially blocked with a plywood barrier to retain a minimum pond depth and ensure that intertidal mesocosms were continuously flooded during the experiment. Five juvenile white shrimp (EGB = 28 – 35 mm total length (TL), SL = 35 – 44 mm TL) were measured to obtain their initial size, individually marked using visible implant elastomer (VIE), and released into each mesocosm to initiate an experiment.

After 6 d the field experiments were terminated. The temporary plywood barrier was removed to allow boat access to the experimental pond. Marked shrimp and other animals were removed from each enclosure using a 2.6-m^2 drop sampler. During recovery of experimental shrimp, only animals on the inside of the mesocosms were collected, and any animals observed

on the outside of these enclosures were released.

In the second approach, we conducted the field experiments in 13-m^2 cages constructed of 1.2-m high walls of 3-mm mesh plastic netting. The vegetated cages extended 2 m into the marsh and tapered from 3 m wide in the marsh to 1 m wide at the opposite end; approximately 6 m^2 of the marsh enclosures were vegetated (Figure 1). The nonvegetated (SNB) cages had the same shape and dimensions but were located between 3 and 8 m from the marsh shoreline. Enclosures were constructed and placed in pairs (one cage of each habitat type) at 6 sites selected randomly at each location (EGB, SL). These experiments, used to estimate growth rates of both shrimp species, were conducted in EGB May 28 – June 3, 2015 and in SL June 19 – 25, 2015. In the EGB experiment, 20 brown shrimp and 10 white shrimp were measured, individually marked with VIE, and released into each enclosure; 20 individuals of each species were used in the SL experiment.

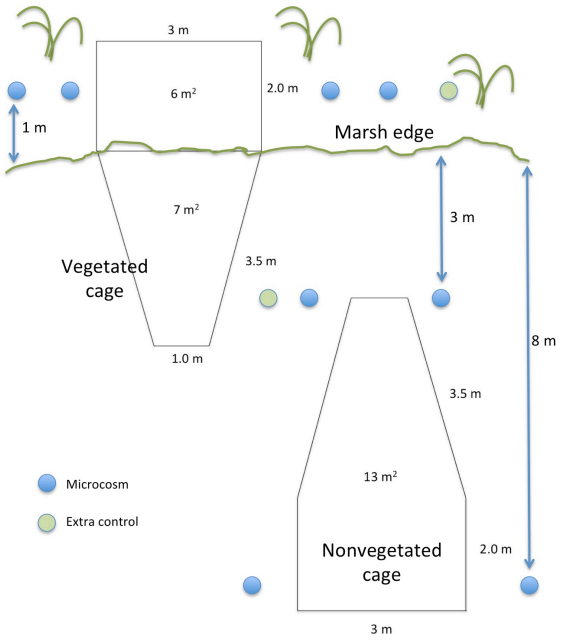

Figure 1. Diagram of a pair of vegetated and nonvegetated cages used in experiments. Filled dots indicate approximate locations of microcosms removed and used in laboratory experiments.

These experiments were terminated after 6 d by seining the inside of each enclosure to recover marked shrimp and other nekton. During the last low tide on the day before the experiment was to be terminated, the vegetated and unvegetated areas within each marsh enclosure was partitioned by placing a wall of plastic netting across the enclosure at the marsh shoreline. Our aim was to prevent shrimp from moving into the marsh during the next high tide when the enclosures would be seined and thereby increase the chance of recovering marked shrimp. Each enclosure was seined at least 7 times and until 3 consecutive sweeps of the seine yielded no marked shrimp.

Experimental shrimp and other nekton collected from the enclosures were immediately placed on ice within labeled sample bags. After all the enclosures were cleared, the marked shrimp from each sample were measured (TL or CL = carapace length) and weighed (mg wet weight), and these data were used to estimate daily growth rates.

Laboratory Experiments

The laboratory experiments were conducted using microcosm cores (20.3 cm diameter, 15 cm deep) extracted at EGB adjacent to the mesocosm sites (two per mesocosm) in 2014 and the cage sites (four marsh and four SNB microcosms per cage) in 2015 (Figure 1) before initiating the field experiments. Ten additional microcosm cores (five in SNB and five in marsh) were collected in 2014 and two extra microcosm cores (one vegetated and one nonvegetated) were collected in 2015 for use as controls in the laboratory experiments. This experimental design provided a total of 50 microcosms (25 vegetated and 25 nonvegetated) for each laboratory

experiment; each microcosm core (0.032 m^2 area) was collected with modified 30-cm high PVC cylinders with 0.5 mm mesh-covered windows for use in the laboratory.

In the Galveston wet lab, the microcosms were placed into three round tanks (1.5 m diameter) connected through a recirculating water filtration system. Vegetated and nonvegetated microcosms were randomly assigned to either fed or non-fed treatments (Table 1). Additionally, 10 microcosms (five vegetated and five nonvegetated) were assigned as controls without experimental shrimp to examine possible effects of microcosm artifacts on infaunal populations.

The external filtration system consisted of a large canister of bio balls, a mesh filter to remove large particulates, and a protein skimmer. Water flow throughout this approximately 2000-liter system was maintained at 266 l min^{-1} with a centrifugal pump, and the temperature was maintained at a level supporting optimal growth (28 – 30 ^oC). The microcosms were placed on concrete blocks within the tanks to maintain a large volume of water in the recirculating system; the bottom of each cylinder was sealed with a plastic plate, and water depth in each microcosm was maintained at approximately 8 cm. *S. alterniflora* stems were clipped flush with the tops of the microcosms. A manifold system was used to divert the recirculating water to each microcosm at 3.8 l min^{-1} through 4.8-mm diameter plastic tubing inserted into the top of each microcosm through a clear plastic lid. Water was discharged from each microcosm through the mesh windows, and this system was designed to ensure a supply of oxygenated water to all microcosms. Light in the lab was provided by artificial fluorescent bulbs on a 12:12 day:night cycle.

Experimental shrimp (white shrimp = 29 – 31 mm TL, brown shrimp = 32 – 35 mm TL), which were collected at the EGB field site and held in the laboratory for 60 h, were tagged with VIE and measured for TL before being placed into the microcosms. One shrimp was used in each of the 40 microcosms (density = 31.2 shrimp m^{-2}). Salinity in the system was maintained at 11 in 2014 and 15 in 2015. Water temperature was measured hourly with HOBO data loggers (Onset Computer Corporation, Bourne, MA). Daily measurements also were made of ammonia, nitrite, and nitrate (API Saltwater Master Liquid Test Kit), dissolved oxygen levels, and salinity (YSI meter) in the system.

We measured growth rates of shrimp in the microcosms over the course of six days. In a similar experiment of longer duration, Whaley (1997) reported growth rates for brown shrimp that were considerably lower than those reported in much of the literature. This may have been due to a decline in infaunal prey density and food limitation over the course of the two-week experiment. With one shrimp in a microcosm, the density is relatively high in comparison to mean natural densities, although such densities have been measured in Galveston Bay marshes (Zimmerman et al. 1984). The short duration of our experiment and including a fed shrimp treatment were designed to address this issue. Shrimp were fed daily with frozen polychaete worms at a rate of about 50% of their estimated body mass. Shrimp size (TL) and wet weights were determined at the end of the experiment (day 6).

Work Completed:

Mean growth rates from our study differed between estuaries (EGB vs. SL), by species (white shrimp vs. brown shrimp), between experimental approaches (field vs. laboratory), and in some cases by habitat type (marsh vs. SNB). The field experiments of 2014 conducted with white shrimp held in SNB provided seemingly valid growth rates, which were within the range of values reported in the literature from other field experiments (Minello & Zimmerman 1991,

Rozas & Minello 2009, Baker & Minello 2010, Rozas & Minello 2011). Mean ± SE growth rates in SNB from these experiments were marginally higher at EGB (1.0 ± 0.114 mm d^{-1} TL and 22 ± 2.164 mg d^{-1}) than SL (0.8 ± 0.034 mm d^{-1} TL and 19 ± 2.133 mg d^{-1}).

These 2014 experiments, however, failed to yield reliable data for shrimp confined to marsh habitat. We recovered few marked shrimp from marsh mesocosms in these experiments (recovery rates: EGB =2%; SL =24%), an indication that experimental artifacts likely contributed to this result. The temporary barrier at EGB failed, and water levels dropped below the depth necessary for shrimp to survive in two of the marsh mesocosms in this experiment. The other marsh mesocosms, however, remained flooded at least 5 cm deep throughout the experiment. Therefore, the failure of the barrier to work properly was not the primary cause of the low recovery or growth rates from marsh mesocosms. Moreover, the barrier at SL performed as designed, and all the mesocosms in this experiment were continuously flooded. Even so, the shrimp in the marsh mesocosms at SL grew slowly (mean ± SE = 0.1 ± 0.056 mm d^{-1} TL) and actually lost weight (mean ± SE = -1 ± 1.299 mg d^{-1}) during this experiment. The dissolved oxygen (DO) concentration measured during the experiment was relatively low in marsh mesocosms, but we obtained valid DO data from only three (two marsh, one SNB) instruments. The most likely explanation for the low recovery and mean growth rate for shrimp in the marsh mesocosms was environmental stress (e.g., low DO, high sulfides) during the experiment. The barrier may have limited tidal exchange and circulation in the pond enough to reduce DO. Alternatively, sinking the mesocosm walls into the marsh soils may have released sulfides or other compounds that degraded the water quality inside the vegetated mesocosms, which reduced shrimp growth. Hydrogen sulfide, for example, is a known toxin to penaeid shrimps and other crustaceans (Kang & Matsuda 1993, Vismann 1996, Nantes & Felder 1998). It seems likely that poor water quality in the marsh mesocosms played a role in this outcome, but the specific cause is not known.

The 2015 field experiments, which incorporated 13 m^2 cages, seemed to work as designed and provided reliable growth data for shrimp in both habitat types. Mean recovery rates were generally higher for brown shrimp than white shrimp and higher at SL than EGB (Table 2). Marked brown shrimp were recovered from all 12 cages of the EGB and SL experiments, but we recovered no marked white shrimp from two cages at EGB and one cage from SL. Although we recovered more shrimp from SNB cages than marsh cages, no significant differences were detected in recovery rates between these habitat types in any of the experiments (Table 2, all p values > 0.15). Mean shrimp growth rates varied by species and habitat types (Table 3). Based on paired t-tests, brown shrimp increased in size (TL and biomass) significantly faster in SNB cages than marsh cages at the SL location. The same analysis detected no significant difference in brown shrimp growth rates between habitat types at EGB. White shrimp growth rates were significantly higher in SNB than marsh, but only at SL when TL was used as the metric for growth in the analysis (Table 3).

The results of the laboratory experiments in 2014 and 2015 were similar. The survival and recovery rates in both experiments were high for shrimp in the SNB treatments (2014 = 100%; 2015 = 100%), but lower in the marsh treatments (2014 = 95%; 2015 = 70%). Growth rates were low in both experiments compared to the values from the field experiments. For example, the mean ± SE growth rate for white shrimp that did not receive additional food in the 2014 experiment was 0.4 ± 0.05 mm d^{-1} TL in marsh microcosms and 0.3 ± 0.07 mm d^{-1} TL in SNB microcosms. The growth rates for brown shrimp in the 2015 experiment were even lower: 0.2 ± 0.09 mm d^{-1} TL in marsh microcosms and 0.2 ± 0.05 mm d^{-1} TL in SNB microcosms. Providing

additional food in these experiments increased shrimp growth rates in both the 2014 (white shrimp: marsh = 0.7 ± 0.07 mm d^{-1} TL, SNB = 0.6 ± 0.08 mm d^{-1} TL) and 2015 (brown shrimp: marsh = 0.4 ± 0.08 mm d^{-1} TL, SNB = 0.4 ± 0.06 mm d^{-1} TL) experiments.

These results provide valuable information on habitat-related growth rates of penaeid shrimps. Our results provide a range of growth rate estimates associated with varying environmental conditions (e.g., salinity, temperature) that can be incorporated into the new SS-3 stock assessment model or used in other models of penaeid shrimp populations.

Applications:

These newly derived growth rates and variance estimates will be directly incorporated into shrimp stock assessment sensitivity model runs. Dependent upon model fits, these rates and variance estimates will be integrated into the assessment models and submitted to the Gulf of Mexico Fisheries Management Council (GMFMC), which must approve any substantive changes to annual stock assessment models. These growth rates will become an integral part of the stock assessments for both brown shrimp and white shrimp once the models are approved by the GMFMC.

Publications/Presentations/Webpages:

References:

Baker, R., and T.J. Minello. 2010. Growth and mortality of juvenile white shrimp *Litopenaeus setiferus* in a marsh pond. Marine Ecology Progress Series 413: 95-104.

Kang, J.C., and O. Matsuda. 1993. Tolerance of anoxia and hydrogen sulfide by benthic crustaceans *Portunus trituberculatus*, *Metapenaeus monoceros* and *Macrobrachium nipponense*. Journal of the Faculty of Applied Science Hiroshima University 32:71-78.

Minello, T.J., and R.J. Zimmerman. 1991. The role of estuarine habitats in regulating growth and survival of juvenile penaeid shrimp. In Frontiers of Shrimp Research, ed. P. DeLoach, W.J. Dougherty and M.A. Davidson, 1-16: Elsevier Science Publishers B. V., Amsterdam.

Nates, S.F., and D.L. Felder. 1998. Impacts of burrowing ghost shrimp, Genus *Lepidophthalmus* Crustacea: Decapoda: Thalassinidea, on penaeid shrimp culture. Journal of the World Aquaculture Society 29: 188-210.

Rozas, L.P. and T.J. Minello. 2009. Using nekton growth as a metric for assessing habitat restoration by marsh terracing. Marine Ecology Progress Series 394:179-193.

Rozas L.P and T.J. Minello. 2011. Variation in shrimp growth rates along an estuarine salinity gradient: implications for river diversions. Journal of Experimental Marine Biology and Ecology 397:196-207.

Vismann, B. 1996. Sulfide species and total sulfide toxicity in the shrimp *Crangon crangon*. Journal of Experimental Marine Biology and Ecology 204: 141-154.

Whaley S.D. 1997. The effects of marsh edge and surface elevation on the distribution of salt marsh infauna and prey availability for nekton predators. MSc thesis, Texas A & M University, College Station, TX

Zimmerman, R.J., T.J. Minello, and G. Zamora, Jr. 1984. Selection of vegetated habitat by brown shrimp, *Penaeus aztecus*, in a Galveston Bay salt marsh. Fishery Bulletin 82:325-336.

Table 1. Design of laboratory experiments showing the number of replicates for vegetated and non-vegetated microcosms in each treatment and the control.

Treatment	Vegetated	Non-vegetated
Shrimp with no food added	10	10
Shrimp with food added daily	10	10
Control (no shrimp and no food added)	5	5

Table 2. Comparison of recovery rates (%) for experimental shrimp held in cages containing two habitat types (Marsh, SNB = shallow nonvegetated bottom). The number of samples (n) from which these means and standard errors (S.E.) were estimated are provided. The dates in 2015 and locations of each field experiment from which these recovery rates were derived also are given. Results of paired t tests (p values) are given for the effect of habitat type on recovery rates.

			Habitat Type						
			Marsh			SNB			
	Date	Location	n	Mean	S.E.	n	Mean	S.E.	*p* value
Brown Shrimp	May 28 - June 3	East Galveston Bay	6	31.7%	3.8%	6	50.6%	10.6%	0.180
	June 19 - 25	Sabine Lake	6	52.5%	12.6%	6	67.5%	9.5%	0.296
White Shrimp	May 28 - June 3	East Galveston Bay	6	28.3%	8.3%	6	41.7%	10.8%	0.158
	June 19 - 25	Sabine Lake	6	40.0%	8.6%	6	53.3%	11.4%	0.357

Table 3. Comparison of growth rates (TL, mm d^{-1} or biomass, mg d^{-1}) for experimental shrimp held in cages containing two habitat types (Marsh, SNB = shallow nonvegetated bottom). The number of samples (n) from which these means and standard errors (S.E.) of growth rates were estimated are provided. The dates in 2015 and locations of each field experiment from which these growth rates were derived also are given. Results of paired t tests (p values) are given for the effect of habitat type on growth rates.

			Habitat Type						
			Marsh			SNB			
	Date	Location	n	Mean	S.E.	n	Mean	S.E.	*p* value
Growth (mm d^{-1})									
Brown Shrimp	May 28 - June 3	East Galveston Bay	6	0.3	(0.040)	6	0.4	(0.040)	0.661
	June 19 - 25	Sabine Lake	6	0.3	(0.021)	6	0.6	(0.021)	0.005
White Shrimp	May 28 - June 3	East Galveston Bay	5	0.5	(0.040)	5	0.6	(0.097)	0.236
	June 19 - 25	Sabine Lake	6	0.4	(0.043)	5	0.7	(0.098)	0.008
Growth (mg d^{-1})									
Brown Shrimp	May 28 - June 3	East Galveston Bay	6	4	(2.060)	6	8	(2.120)	0.239
	June 19 - 25	Sabine Lake	6	5	(1.815)	6	24	(3.351)	0.003
White Shrimp	May 28 - June 3	East Galveston Bay	5	16	(3.855)	5	23	(4.057)	0.099
	June 19 - 25	Sabine Lake	6	32	(5.443)	5	32	(6.719)	0.871